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(54) Title: COMPOSITION CONTAINING CREATINE AND PHOSPHORUS

(57) Abstract: The invention discloses a composition comprising (a) soluble creatine, (b) a phosphorus supplement, wherein the phosphorus supplement provides at least 75 % of the recommended daily dose of phosphorus value per serving, and (c) a blood buffer, which is preferably citrate and/or (bi)carbonate. The composition is used in a method for increasing the energy capacity within tissue cells.

Composition containing creatine and phosphorus

FIELD OF THE INVENTION

The invention pertains to nutritional compositions containing creatine and phosphorus, and to methods of increasing the energy capacity and, in particular the anaerobic working capacity of humans and animals by administering such compositions.

BACKGROUND OF THE INVENTION

The energy level of cells is dependent on numerous factors, often referred to as the cell metabolic state. Metabolism can be defined as the sum of all chemical reactions occurring in a cell, many of which are for breaking down nutrients, many of which are for building other molecules. Metabolism involves both catabolism and anabolism.

Catabolism is the decomposition of complex molecules to more simple molecules. It is performed by cell enzymes in order to extract covalent bond energy. The extracted energy is then used to perform anabolism, wherein energy derived from catabolic processes is converted to high energy phosphate bonds of for example adenosine triphosphate (ATP).

During high energy requiring activities, especially within muscle cells, the ATP pool is rapidly depleted and ATP has to be quickly resynthesized. It is believed that during highly intensive exercise for a limited period of time, ATP regeneration from ADP is for a significant part accomplished by the anaerobic degradation of phosphocreatine. Maximal anaerobic work is therefore believed to be limited by the breakdown of ATP and phosphocreatine (7). In line with this hypothesis, it has been demonstrated that high intensity exercise resulted in a 35 to 96 % depletion of muscle phosphocreatine levels, depending upon the duration of the activity (13,14,19).

A number of studies have shown that anaerobic working capacity (AWC) from the critical power test provides a theoretically and experimentally valid estimate of work capacity associated with muscle energy reserves (4,15,16). The muscle energy reserves are formed by the ATP and phosphocreatine pools present within the muscle cells. These reserves are especially important for subjects involved in intermittent and non-endurance activities, such

as intermittent and non-endurance sports. Clear examples of such activities include for example athletics, field sports, cycling, running, bodybuilding, etc.

Creatine, the precursor of phosphocreatine, is an amino acid (*N*-amidino-*N*-methylglycine) that is found in many foods and feeds and is an important regulator in the energy supply and energy storage within the cell. Creatine is synthesized from glycine, arginine and methionine in the liver, pancreas and kidney and subsequently released into the blood stream. The cellular concentration of creatine is determined by the ability of the muscle cells to take up creatine, because muscle cells cannot synthesize creatine. Phosphocreatine is generated by conversion of creatine to phosphocreatine by creatine kinase. The concentration of phosphocreatine in the muscle is believed to be about 4 times greater than that of ATP and is about 3 times greater than that of creatine.

Increased capacity of muscle cells to perform the energy consuming muscle contraction will increase performance. Therefore subjects involved in high energy consuming activities often are administered nutritional supplements comprising compounds involved in cell metabolism, to enhance performance. For example, the (oral) administration of creatine-containing supplements is widely accepted by subjects involved in high energy consuming activities such as athletes and animals and is believed to improve performance by increasing intracellular muscle phosphocreatine/creatine pools.

A number of studies have demonstrated that a daily intake of creatine monohydrate 4×5 gram creatine per day, also referred to as creatine loading, for five to six days can significantly increase skeletal muscle creatine content by an average of 20%, with phosphocreatine accounting for 20% of the increase (12,19). Additionally, several studies have demonstrated ergogenic benefits of creatine monohydrate loading on isokinetic strength (10), running (3), cycling (1,2,5,11), jumping (3,19) and bench-press (19) performance. Furthermore, Casey et al. (5) reported a significant correlation ($r = 0.72$, $p \leq 0.05$) between anaerobic performance during isokinetic cycling and skeletal muscle creatine retention from creatine monohydrate loading.

WO 98/06278 describes methods and compositions for increasing the anaerobic working capacity in tissues and provides a method for increasing the synthesis and accumulation of

beta-alanylhistidine dipeptides, with a simultaneous increase in the accumulation of creatine, in bodily tissues of humans and animals. This is accomplished by causing an increase in the blood plasma concentrations of beta-alanine and creatine, or the blood plasma concentrations of beta-alanine, L-histidine and creatine, by the ingestion or infusion of a composition including beta-alanine, beta-alanine and creatine, or beta-alanine, L-histidine and creatine, or active derivatives thereof.

WO 00/30634 relates to the use of creatine or a creatine analogue for reducing the biological process of aging, for treating disuse muscle atrophy or for stimulating subsequent restoration of muscle mass in rehabilitation training. The muscle atrophy is the result of immobilization, or reduced level of physical activity due to either disease or aging. The biological process of aging comprises for example the simultaneous degeneration and degradation of body tissues, including muscle and bone, neural tissue and secretory cells. This document further relates to a therapeutic preparation for treating or preventing disuse muscle atrophy and for inhibiting the biological process of aging, comprising a suitable carrier, diluent or excipient and an effective amount of one or more creatine compounds

The exposure of creatine, such as creatine monohydrate, to the acidic environment of the stomach is believed to result in an irreversible formation of creatinine from the creatine. Since creatinine has no function in the cell, creatinine is excreted.

US patent 5,908,864 provides a creatine supplement which does not quickly convert to creatinine and is easily and conveniently ingested by a person. It discloses a nutritional gel containing creatine and the method of producing the creatine gel. The creatine gel is made by crosslinking maltodextrin and a modified starch through an aqueous endothermic reaction at approximately 90°C. A buffering agent, such as potassium phosphate, is added to the gel to maintain a pH value at approximately 7.0. The gel is then cooled and creatine is added. Next, the gel is stabilized bacteriologically by adding a preservative, such as potassium sorbate to the gel.

US patent 5,973,199 provides hydrosoluble organic salts of creatine. The salts are useful in the dietetic and food industry.

US 5,925,378 provides a method for enhancing a stable concentration of cellular creatine in a human. A mixture containing an effervescent and an acidic edible salt form of creatine is completely dissolved in water. The resulting the solution is immediately ingested, and an effective amount of creatine is absorbed. Preferably, the effervescent is in the form of a tablet which contains creatine in the form of an edible salt, a mixture of acids, and sodium.

Besides creatine, phosphate and other phosphorus compounds play an important role in energetic capacity within the body. Phosphate plays a vital role in ATP resynthesis and phosphocreatine synthesis, since it is a substrate in the phosphorylation of ADP and creatine. Inorganic phosphate (Pi) supplementation is believed to provide a number of ergogenic benefits. It has been reported to increase aerobic capacity by increasing 2,3-diphospho-glycerate (DPG) levels in red blood cells (Cade et al., Med Sci Sports Exerc 16(3):263-8, 1984; Kredier et al., Med Sci Sports Exerc 22(2):250-6, 1990). The principal role of 2,3-DPG is to facilitate oxygen release from the binding of haemoglobin so that the oxygen can diffuse faster into working muscle cells.

Several other mechanisms have been proposed through which phosphate supplementation may affect performance, besides increase of 2,3-DPG levels. These include the promotion of phosphate-stimulated glycolysis, increased availability of phosphate for oxidative phosphorylation and creatine phosphate synthesis, an increased red cell anaerobic glycolytic efficiency, and enhanced myocardial and cardiovascular efficiency. Although theoretical rationales for phosphate supplementation for ergogenic benefits are known in the art, the ergogenic benefits of phosphate supplementation have not been consistently observed and little scientific evidence exists to recommend exogenous phosphate as an ergogenic aid (21).

US 5,968,544 discloses an acidic composition for human consumption, comprising creatine. The composition is conveniently an isotonic drink for storage at 4°C or a dry stable powder which may be stored at ambient temperature. Optionally included in the composition are minerals in a limited quantity; e.g. typical amounts of phosphates included are 50 mg/liter.

Recently, Wallace et al. (20) studied the effect of the separate and combined short term creatine monohydrate and sodium phosphate supplementation on the body composition, performance, and blood chemistry. Subjects were randomly assigned to ingest either 1000

mg of tribasic sodium phosphate (P), 5000 mg creatine monohydrate (CR), 5000 mg creatine monohydrate and 1000 mg tribasic sodium phosphate, or a glucose placebo (G) four times daily for 5 days. Results indicated that following the phosphate trials, VO2 max (8%) and the ventilatory anaerobic threshold were significantly elevated (10%). In contrast, both creatine monohydrate loading trials resulted in a significant increase in body mass (1.35 ± 0.22 kg, mean SE) with no change in fat free mass ($p<0.05$). The only significant increase observed for concurrent creatine and phosphate loading compared to individual phosphate or creatine loading was an elevated power output suggesting that there may be some performance benefit to short-term creatine and phosphate loading. However, the effect of concurrent creatine and phosphate loading (6% increase in power output compared to placebo) is a mere additional effect of the phosphate (3% increase in power output compared to placebo) and creatine loading (3% increase in power output compared to placebo).

US 5,973,005 discloses a stable aqueous solution of creatine acid sulfate providing a source of creatine to an animal when taken orally. The aqueous solution of creatine acid sulfate (after neutralization and buffering) has a pH of about 7.2 to about 7.8 and is stable for at least six months at room temperature. The creatine acid sulfate is produced by adding creatine monohydrate to a sulfuric acid solution in a stoichiometric amount to result in creatine acid sulfate having a pH initially of 2.0-3.0. The resulting solution is diluted with water and neutralized to raise the pH and avoid the formation of creatinine. The solution preferably contains a buffering and neutralizing agent such as tribasic potassium phosphate which forms mono- and dibasic potassium phosphates by interaction with the hydrogen ions liberated from the acid sulfate. The aqueous solution can be combined with a sweetener, electrolyte and carbohydrate source to produce a stable drink for providing a source of creatine to an animal in need thereof. An effective amount of glycerol is preferably added to enhance absorption of the creatine through the intestinal wall into the bloodstream and eventually to the needy skeletal muscles.

US 5,397,786 discloses a liquid composition to be used as a rehydration drink, particularly suited for people who do heavy work under severe conditions, e.g. at high temperatures, and for sports people and athletes, as well as for patients who exhibit dehydration symptoms due to severe illnesses such as diarrhea or vomiting, contains per serving unit water at least 1 to

100 g of at least one carbohydrate, such as glucose polymers, maltodextrin and fructose; 2 to 2500 mg of at least one electrolyte, such as an alkali and/or earth alkali salt; 0,1 to 750 mg of at least one ammonia neutralizer, such as D,L-magnesium aspartate, L-arginine and glutamate; at least one energy enhancer, such as members of the vitamin B group and branched chain amino acids; at least one antioxidant such as beta-carotene, vitamin C, vitamin E and selenium; 1 to 30 mg of at least one membrane stabilizer, such as choline chloride, betaine chloride and methionine; and 1 to 200 μg of at least one neuromuscular function enhancer such as octacosanol.

During high intensity exercise, the anaerobic conversion of glucose, i.e. glycolysis, results in the formation of hydrogen ions (H^+), inducing chemical changes, e.g. a decrease of blood pH. Decrease of blood pH results in for example pain and leads to decreased performance of muscles. Blood buffers in the blood protect against large changes in pH. It is believed within the art that suitable buffering substances, when taken orally, can increase performance.

The energy requirement for the covalent bonding of phosphate to ADP is very often generated in the Krebs cycle. The Krebs cycle involves a series of enzymatically controlled reactions enabling complete oxidation of two-carbon acetyl groups to water and carbon dioxide. Each complete turn of the Krebs cycle yields one molecule of ATP, 3 molecules of NADH and one molecule of FADH2. The latter are subsequently used as electron donors in the electron transport system to yield additional ATP.

WO 99/21565 describes a pharmaceutical composition which includes a sugar and a Krebs cycle intermediate, or salt thereof, or a precursor of a Krebs cycle intermediate. Krebs cycle intermediates include citric acid, aconitic acid, isocitric acid, alpha-ketoglutaric, succinic acid, fumaric acid, malic acid, and oxaloacetic acid, and mixtures thereof. Precursors of Krebs cycle intermediates are compounds converted by the body to form a Krebs cycle intermediate. The pharmaceutical composition is described to be particularly desirable for the prophylaxis or treatment of disorders associated with impaired mitochondrial function. Disorders that can be treated include conditions or diseases characterized by a decreased level of oxidative metabolism, such as conditions or diseases of the nervous system, conditions or diseases of other parts of the body and conditions or diseases of the body as a whole. The pharmaceutical composition described can also include an adjuvant for enhancing

mitochondrial function (i.e., oxidative metabolism). Suitable adjuvant include vitamins, minerals, antioxidants, and other metabolism-enhancing compounds. Exemplary metabolism-enhancing compounds include L-carnitine and its derivatives, and creatine.

Notwithstanding the above disclosures, there remains a need in the art for compositions, which increase the capacity of energy output from cells. Especially, compositions suitable for enhancing body strength or performance in intermittent and non-endurance sports, for example prevention, extension and treatment of muscle fatigue especially during high energy requiring activities for a relatively short period of time are desired. The composition should solve problems with respect to insufficient power output, especially insufficient short-term power output. The composition should also increase power output, and especially increase anaerobic working capacity of subjects in need thereof.

SUMMARY OF THE INVENTION

The present invention provides desired and novel compositions, which increase the capacity of energy output from cells, suitable for optimizing performance, especially muscle performance, and/or for enhancing muscle cell recovery after exercise.

The invention provides a composition, which shows surprisingly synergistic effects of creatine, phosphorus and blood buffers. Without being bound by theory, it is the inventors' believe that this synergistic effect is caused by the simultaneous increase of cellular phospho-creatine and ATP pool, increase of glycolytic capacity and counteracting the adverse side effects of increased glycolytic capacity. These concurrent actions result in a significant increase in anaerobic working capacity, providing cells, especially muscle cells, the opportunity to increase energy output and thus the ability to perform muscle contraction.

The invention further pertains to compositions for increasing the anaerobic working capacity of a subject, making the composition suitable for subjects in need of increased performance, for example by remaining performance at a high level for a longer period of time.

DETAILED DESCRIPTION OF THE INVENTION

The composition according to the invention is especially suitable for the subjects in need of an increased energy capacity within tissue cells, for example an increased power output

and/or high power output for a longer period of time. Subjects wishing or requiring enhanced performance of the body such as an increased capacity to perform muscle contractions, or increased power output for a relatively short period of time can use the composition according to the invention advantageously. Furthermore, the composition according to the invention can be used in a method for treatment or prevention of disorders, diseases or abnormal physical states, especially associated with energy levels within the cell.

The composition was shown to be especially suitable for increasing the anaerobic working capacity (AWC) of a subject. AWC can be defined as the capacity to extract energy from phosphocreatine/ATP pools and/or glycolysis. Glycolysis is defined as the ATP generation via conversion of glucose or glycogen to lactate. An enhancement of the AWC accomplished by the composition according to the invention will for example result in: a) the ability to increase "explosive" power output, i.e. exercise wherein energy is mainly extracted from glycolysis and ATP/phosphocreatine pool; b) an extended exercise length; c) increased performance during competition; d) increased ability to recover from high intensity activities or e) increased performance in other activities with high energy requirements over a relatively short time period. Subjects wishing or requiring an increased AWC are often preparing for or practicing intermittent and non-endurance sports.

It was unexpected that the AWC would increase by administration of the composition according to the invention, although it has been suggested in the art that phosphate supplementation could increase the effects of creatine loading. AWC increase has been reported as an effect of creatine administration, however, phosphorus supplementation is believed in the art to increase aerobic capacity, via stimulation of 2-DPG formation. Results according to Wallace et al (20) indeed suggest that the effect of concurrent phosphate and creatine supplementation on power output is merely based on an additional effect and not a synergistic effect. Table 1 shows that the concurrent administration of just creatine and phosphate has a mere additional effect on the power output (results from Wallace et al).

Furthermore, it was shown that the effect on AWC achieved with a composition according to this invention is not a mere additional effect of phosphate, creatine and blood buffer supplementation, since the combination creatine/blood buffer only caused a 16% increase in anaerobic working capacity, while with composition according to the invention an increase of

about 50% was observed. Although not wishing to be bound by theory, the inventors believe that the increased effect is caused by the combination of phosphorus, creatine and blood buffer, wherein creatine and phosphorus provide substrates for the intracellular creatine/phosphocreatine pool, wherein the phosphorus supplement increases glycolysis and wherein the blood buffer counteracts the effect of an increased glycolysis caused by the phosphorus supplementation. As a direct consequence of reduced adverse effects of increased glycolysis, glycolysis reactions and other reactions required for muscle contractions will proceed more efficiently and/or for a longer period of time, compared to a situation wherein the adverse effects of increased glycolysis are not counteracted. This results in an increased ability of cells to provide energy.

Table 1: Increase of power output and anaerobic working capacity compared to placebo

	Power output according to Wallace et al (20)	AWC using composition of the invention
Placebo	-	-
Creatine	3 %	nd
Phosphate	3 %	nd
Creatine + Blood buffer	nd	15.6%
Creatine + Phosphate	6 %	nd
Creatine + Phosphate + Blood Buffer	nd	49.8 %

The composition according to the invention is also especially useful for subjects who experience or need to be prevented from a decrease or insufficient increase in power output and/or anaerobic working capacity. The composition according to the invention can thus also be advantageously used prior to, during or after for example muscle disuse, e.g. due to partial or complete immobilized of the body, e.g. due to hospitalization, subjects spending a significant time in an environment with reduced gravitation such as astronauts; etc. Use of the composition in such events will prevent a large drop-back of muscle quality and facilitate recovery.

The creatine used in the composition according to the invention can be any creatine known in the art, including for example creatine monohydrate, creatine phosphate, creatine sulfate. Preferably, however, creatine comprises a creatine salt consisting of anionic and cationic part, wherein the cationic part consists of and provides creatine or creatine analogs or derivatives.

Preferably a highly hydrosoluble salt of creatine is used as a source of creatine, since it is believed that a high solubility of creatine facilitates the absorption of creatine by the intestines and ultimately results in significantly higher increase of the intracellular creatine and phosphocreatine concentrations. Preferred hydrosoluble creatine salts are those having a solubility above about 6 grams of the salt per 100 ml water of 37°C, preferably above about 7.5 grams of the salt per 100 ml water, even more preferably above about 10 grams of the salt per 100 ml water of about 37°C. The solubility of the creatine salt used in the composition of the invention preferably has a limited time required to dissolve the organic salt of creatine.

The creatine salt to be used is especially a highly soluble organic creatine salt. Organic creatine salts are those creatine salts wherein both the anionic and cationic part of the salt are organic. The organic anionic part or the organic creatine salt can serve important functions within the subjects body, without the need for addition of a specific additional component or composition, which can make the composition unnecessarily complex and/or expensive. Preferred anions are those derived from (aliphatic) polycarboxylic, especially dicarboxylic and tricarboxylic organic acids, preferably having 3-6 carbon atoms, optionally containing 1-2 carbon-carbon double bonds, 1-4 hydroxyl groups and/or 1-2 keto groups, including citrate, maleate, fumarate, tartrate, succinate, oxaloacetate, malate, and gluconate. According to a particularly preferred embodiment, the anionic part of the creatine salts is capable of providing a blood buffer, for example citrate, or a Krebs cycle intermediate or precursor thereof, for example malate, fumarate or citrate. Especially preferred creatine salts include those wherein the anion provides both a blood buffer and a Krebs cycle intermediate, for example creatine citrate. According to a preferred embodiment, a creatine salt is used having citrate as the anionic part and protonated creatine as the cationic part of the salt. Both creatine, dicreatine and, where applicable, tricreatine salts of the polycarboxylic acids are suitable, and wherever creatine citrate is used in this disclosure, this can be replaced by dicreatine citrate or tricreatine citrate and vice versa. Tricreatine citrate is preferred.

The quantity of creatine provided per weight of creatine salt depends on the type (molecular weight) of the creatine salt used. Preferably the composition according to the invention provides about 0.01 to about 1, preferably about 0.02 to about 0.5, most preferred about 0.04 to about 0.25 grams creatine per kilogram body weight per serving. For humans beings the

composition would provide about 0.5-100 grams creatine per serving, more preferably about 1-50 grams, most preferred about 2-25 grams, for example 2.5 – 10 gram, e.g. 5 ± 0.5 g.

Without being bound by theory, the inventors believe that the phosphorus supplement within the composition according to the invention increases the ability of cells to provide energy via the glycolysis. During glycolysis, glucose or glycogen is first converted to glucose-6-phosphate, requiring phosphorus, and ultimately to pyruvate. Pyruvate can serve as a substrate within the Krebs cycle (aerobic) or results in the formation of lactate and hydrogen ions. The term glycolysis whenever used in the context of this disclosure refers to the conversion of glucose into lactate and hydrogen ions. Additionally, the phosphorus may serve as a substrate for the formation of ATP/phosphocreatine pool.

The recommended daily intake of phosphorus for humans differs in each country and is depending on age and sexes. According to the Nordic Nutrition Recommendations of 1996 (well known to those skilled in the art), about 700 mg phosphorus should be ingested by a young human on a daily basis and about 600 mg phosphorus is recommended for humans above 22 years. However, the US recommended daily allowances (RDA) is 700 mg for the group above 22 years. For clarity, whenever referred to a daily dose value for humans in the disclosure of this invention, this daily dose will refer to 700 mg phosphorus for a human being. The use of this value does not limit the scope of this invention, if alternative values for recommended doses are used. Mere recalculation based on the weight of phosphorus used in the composition according to the invention will provide the percentage of phosphorus based on the alternative recommended daily intake for phosphorus. For non-human mammals, the amounts to be used can be calculated in an analogous manner, on the basis of the recommended or usual daily intakes and the weight of the animal. The term phosphorus supplement includes any supplement, organic or inorganic, wherein a significant amount of phosphorus is present. Preferably the phosphorus is not part of the creatine (salt). Where amounts of phosphorus are given, these refer to elemental phosphorus (P).

The phosphorus supplement included in the composition according to the invention should provide in at least about 75% of the daily dose value for phosphorus per serving, preferably above about 90 %, even more preferred above about 100%, especially preferred above about 110%, for example above about 125%. The composition should not exceed about 25 times

the daily dose value of phosphorus per serving, preferably it comprises below about 15 times the daily dose value. For humans a daily dose of the composition would thus preferably provide above about 500 mg phosphorus per serving, preferably above about 600 mg phosphorus, especially above about 700 mg phosphorus, for example 1000 ± 100 mg phosphorus, however it should not exceed 17.5 g, preferably it should be below about 10 g.

Preferably the composition according to the invention has a weight ratio of phosphorus to creatine of about 1:25 to about 10:1, preferably about 1:10 to about 1: 1, most preferably about 1:6 to about 1:4. Especially preferred are those composition providing both at least about 75% of the recommended daily dose of phosphorus and have a phosphorus to creatine ratio of about 1:25 to about 10:1.

Suitable phosphorus containing supplements include those supplements comprising bio-available phosphorus, for example salts including phosphate. According to a preferred embodiment, inorganic salts including phosphates, for example sodium phosphate, potassium phosphate are used. According to a preferred embodiment a mixture comprising sodium phosphate and potassium phosphate is used, wherein the potassium: sodium ratio is between 5:1 and 1:5 on a molar basis. According to a further preferred embodiment, monobasic phosphate salts are used as a source of phosphorus.

Without being bound by theory, the inventors believe that the blood buffer is required in the composition according to the invention since it inhibits the undesired side effects of increased glycolysis, such as blood pH decrease. The term blood buffer includes all such compositions recognized in the art as suitable blood buffers. Examples of such buffers include salts comprising carbonate, such as sodium bicarbonate, potassium bicarbonate, sodium carbonate, potassium carbonate or mixtures including one of these compounds or citrate or citric acid. Unsuitable for use as a blood buffer is phosphate, since phosphate buffers fluids within the body such as urine. Furthermore sulfate is considered as unsuitable, as it provides insufficient, if any, blood buffering capacity and the smell and taste of sulfate are not appealing.

Since a high intake of vast amounts of carbonate has several negative side effects, such as explosive diarrhea, according to a preferred embodiment, the blood buffer comprises only a

limited amount of carbonate or bicarbonate, in combination with a second suitable blood buffer, for example citrate and/or citric acid. According to a preferred embodiment, the composition according to the invention comprises a weight ratio buffer : creatine of about 1:50 to about 50:1, preferably 1:10 to 10:1, even more preferred 5:1 to 1:5, e.g. about 1:1. Suitable amounts of carbonate and/or bicarbonate include above about 10 mg per gram of creatine, preferably above about 100 mg per g of creatine, more preferred above about 250 mg per g creatine. Suitable amounts of citric acid include above about 10 mg per gram of creatine, preferably above about 100 mg per gram of creatine, more preferred above about 250 mg per gram creatine. Suitable amounts of citrate are above about 10 mg per gram of creatine, preferably above about 100 mg per gram of creatine, more preferred above about 200 mg per gram creatine.

According to a further preferred embodiment the composition according to the invention comprises one or more Krebs cycle intermediates or precursors thereof. Krebs cycle intermediates are the acids utilized in or during the Krebs cycle. Thus, Krebs cycle intermediates include citric, aconitic, isocitric, succinic, fumaric, malic and oxaloacetic acid. Precursors of Krebs cycle intermediates are those which upon administration to a subject are converted by the body into a Krebs cycle intermediate. Precursors of Krebs cycle intermediates include mono- and di-alkyl citrates, aconitates, isocitrates, α-ketoglutarates, succinates, fumarates, malates, and oxaloacetates and are especially suitable. Preferably citrates, fumarates, malates or mixtures including one of the previous are included. Suitable sources for Krebs cycle intermediates or precursors thereof include salts including precursors of Krebs cycle intermediates, for example sodium citrate or Krebs cycle intermediates such as for example citric acid. According to a particularly preferred embodiment the Krebs cycle precursor is provided by the anionic part of the creatine salt.

The composition according to the invention advantageously further comprises an adjuvant for enhancing mitochondrial function, e.g. the oxidative metabolism. Suitable adjuvants include vitamins and minerals, for example vitamin B, especially vitamin B6, vitamin B1 and/or vitamin B2. The composition according to the invention can also include an adjuvant for enhancing the cells metabolism. Such compounds include L-carnitine and carbohydrates.

Carbohydrates included within the composition according to the invention can be any carbohydrate known in the art, suitable and able to ultimately function as a Krebs cycle substrate, or a pentose such as ribose. Such carbohydrates thus include digestible monosaccharides and polysaccharides. Preferred monosaccharides include glucose and fructose. Preferred polysaccharides include sucrose, lactose, maltose, starch and starch fractions and hydrolysates. Non-digestible carbohydrates (nutritional fibres) may also be present.

The quantity of digestible carbohydrates included in the composition of the invention greatly depends on the purpose and subject to which the composition according to the invention is administered. For example, the composition can be admixed to feed or food having a significant carbohydrate content. For use by humans, the composition of the invention preferably provides about 0.5 to about 500 grams of carbohydrates, more preferably about 1 to 200 grams, most preferably about 5 – 100 grams on a daily basis, for example 5-15, especially about 12 grams per serving.

The composition according to the invention further advantageously comprises compound or mixture of compounds having an effervescent action. Creatine may thereby be uniformly and accurately dispensed when completely dissolved in liquid. Suitable compounds include sodium bicarbonate, potassium bicarbonate, sodium carbonate, potassium carbonate or mixtures including one of these compounds. The inclusion of an effervescent will facilitate ease of usage of the composition, since it will decrease preparation time and facilitate proper mixing of the composition according to the invention with water. According to a preferred embodiment, the composition used as an effervescent additionally has the capacity to provide blood buffering capacity.

The composition according to the invention can further comprise components suitable for providing taste or colour. Exemplary taste modifiers include artificial or natural flavorings such as citric flavours, e.g. fruit punch having lemon, orange or grapefruit and artificial sweeteners such as sucralose, aspartame or saccharin. Colouring can be provided by e.g. beta-carotene.

An especially effective method for increasing the anaerobic working capacity comprises a building phase and a maintenance phase. During the building phase the composition

according to the invention is administered 1.5-10 times the quantity required in the maintenance phase, preferably about 2.5-6 times. The building phase generally requires about 2-15 days for a human, for example about 4-9 days. Preferably about 4 servings per day are administered to a subject during the building phase and about 1 serving per is administered to a subject during the maintenance phase. Alternatively, servings with about 4 times lowered levels of the active components can be served during the maintenance phase. For optimum results the subject is subjected to physical exercise during or after the building phase and during the maintenance phase.

The composition according to the invention can be advantageously used by human subjects. Especially human males can use the composition advantageously. Although human female subjects showed a 10.8 % increase in AWC after 6 days supplementation compared to placebo, human males showed a 49.8 % increase in AWC after 6 days supplementation compared to placebo.

The composition according to the invention is preferably administered orally as a nutritional supplement. The composition can for example be added to food or feed products. Preferably the composition is provided in an edible bar or cookie, a soluble powder, drink, tablets or capsule. A powder, to be reconstituted in an aqueous liquid, is preferred. In order to ensure easy administration of the reconstituted powder, in particular if an effervescent is present, the mass of dry matter of the liquid/powder should be below 0.1 g per ml, preferably below 0.08 g/ml, even more preferably below 0.07 g/ml. The mass of powder to be reconstituted in water is preferably limited, so that the consumption of large quantities can be avoided. Preferably the mass of powder per serving does not exceed 50 grams, more preferably it is between 20 and 35 grams. In order to ensure sufficiently solubilised creatine, the concentration of creatine in the reconstituted product is preferably between 0.5 and 1.5 gram creatine/100 ml. The daily dose may administered through a single unit (tablet, bar, cookie, capsule, sachet, etc.) or through multiple daily units, e.g. 2, 3, 4 or 6 units, where the total of the daily units correspond to the amounts given herein as daily dose.

EXAMPLE 1: *Sports trial*

Thirty-one healthy men were tested.

Following pre-testing, the subjects were randomly assigned to one of three treatment

conditions using a double blind design:

1) 20 g of flavoured dextrose (CHO) plus blood buffer in the form of effervescent carbonate/ bicarbonate powder as a placebo (PL);

2) 18 g CHO, blood buffer and 5 g creatine, the blood buffer and the creatine being provided in the form of creatine citrate, and effervescent carbonate/bicarbonate powder in a flavoured effervescent powder blend (Creatine Edge Effervescent; Fortress Systems, Omaha) (Cr);

3) 18 g CHO, blood buffer, 5 g creatine and 1 g phosphorus, the blood buffer and the creatine being provided in the form of creatine citrate, and effervescent carbonate-/bicarbonate powder in a flavoured effervescent powder blend (Creatine Edge Effervescent; Fortress Systems, Omaha) (CP).

The subjects ingested the supplements four times per day for two and six consecutive days before returning to the lab for post-testing.

Critical Power Test

Thirty healthy men completed three phases of testing on a calibrated electronically braked cycle ergometer (Corival 400, Quinton Instruments).

1) pre-testing consisted of two bouts (= exercise session) performed at power outputs selected to elicit fatigue in 1 to 10 minutes;

2) post-testing following two days of supplementation required two work bouts at power outputs identical to those performed during pre-testing.

3) post-testing following six days of supplementation required two work bouts at power outputs identical to those performed during pre-testing.

The procedure for the critical power (CP) test is the same as previously described by Moritani et al. (16). The power outputs for the exercise bouts ranged from 200 to 375 watts (W) depending upon the fitness level of the subject. Rest periods between each exercise bout was continued until the subjects' heart rate returned to within 10 beats per minute of pre-exercise levels (this usually takes 30 minutes or longer; 20-21). The subjects performed both exercise bouts on one day.

Prior to each exercise bout, the seat height of the cycle ergometer was adjusted for near full extension of the subject's legs while pedalling and foot straps were adjusted to prevent the feet from slipping off the pedals during testing. Subject did warm up for four minutes by pedalling at a power output of 30 W. Following a 2-minute rest period, the subjects began

pedalling against zero resistance and, upon reaching a pedalling rate of 70 rpm, the appropriate power output was applied within the first 2-3 seconds of the test. Each subject was encouraged to maintain the required pedalling rate throughout the entire exercise bout. The exercise bout was immediately terminated when the subject was unable to maintain 65 rpm as determined by the rpm monitor on the ergometer. Time limit (TL) was recorded to the nearest 0.1 second.

Work limit (WL) was calculated by multiplying power (P) and TL (WL = P X TL). Anaerobic working capacity (AWC) is the amount of work in kilojoules corresponding to the Y intercept of the WL - TL relationship as previously described (15-16). Test-retest reliability data for AWC (anaerobic working capacity) for young adult males (n = 11) measured 7 days apart resulted in an intraclass correlation (R) of 0.97 with a SEM of 0.59 kJ.

Results

Table 2: Mean AWC (kJ) for each treatment group before treatment (0 days) after 2 days and after 6 days of supplementation. % change represents the average increase in mean AWC after 6 days supplementation.

Table 2

Group	n	0 days	2 days	6 days	% change
Blood buffer	10	16.2	13.6	15.7	0 %
Creatine+buffer	10	17.5	16.7	20.2	15.6 %
Creatine+buffer+P	11	15.0	18.5	22.4	49.8 %

It was surprisingly found that the composition according to the invention resulted in a much greater increase in AWC than the ingestion of creatine without phosphate.

EXAMPLE 2: *Soluble powder*

A powder to be reconstituted in water or fruit juice comprising:

7.7 grams tricreatine citrate (provides 5 gram creatine)
12 grams fructose
2.06 grams monobasic potassium phosphate
2.06 grams monobasic sodium phosphate
3.8 grams sodium bicarbonate
150 mg sodium carbonate

4.6 grams citric acid
artificial/natural fruit punch
artificial colours.

EXAMPLE 3: *Solubility*

Using HPLC, the saturated solubility of dicreatine citrate and creatine monohydrate in water of 37°C was determined to be 148.1 (± 0.7) and 57.6 (± 9.3), respectively. Thus the saturated solubility of dicreatine citrate is about 2.6 times higher than that of creatine monohydrate.

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Claims

1. A composition comprising:
   a. creatine;
   b. a phosphorus supplement, wherein the phosphorus supplement provides at least 75% of the recommended daily dose of phosphorus value per serving;
   c. a blood buffer.

2. A composition according to claim 1, wherein the weight ratio of phosphorus to creatine is about 1:25 to about 10:1.

3. A composition according to claim 1, wherein the weight ratio of phosphorus to creatine is about 1:10 to about 1: 1, preferably about 1:6 to about 1:4.

4. A composition according to any one of claims 1-3, wherein the phosphorus supplement comprises an inorganic salt comprising phosphorus.

5. A composition according to any one of claims 1-4, wherein the creatine is a creatine salt.

6. A composition according to claim 5, wherein the creatine is an organic creatine salt.

7. A composition according to claim 6, wherein the creatine salt comprises an anionic component selected from the group of tartrate, maleate, malate, fumarate, citrate, and pyruvate.

8. A composition according to any one of claims 5-7, wherein the creatine salt has a solubility above 6 grams per 100 ml water.

9. A composition according to any one of claims 1-8, wherein the blood buffer is selected from the group consisting of carbonate, bicarbonate, citrate and citric acid.

10. A composition according to any one of claims 1-9, further comprising a Krebs cycle intermediate or precursor thereof.

11. A composition according to claim 10, wherein the creatine is an organic creatine salt and the anionic component of the creatine salt is a precursor of a Krebs cycle intermediate.

12. A composition according to any one of claims 1-11, further comprising carbohydrate.

13. A composition according to any one of claims 1-12, comprising 1-10 gram creatine, preferably provided by creatine citrate, 0.6 – 5 gram phosphorus, preferably provided by phosphate, 0.1 – 15 gram buffer, preferably a combination of carbonate and/or bicarbonate and citrate, and 1-100 g of digestible carbohydrates.

14. A composition according to any one of claims 1-13, further comprising an effervescent.

15. A composition according to any one of claims 1-14, further comprising a pentose, preferably ribose.

16. A composition according to any one of claims 1-15, further comprising a sodium salt, preferably sodium phosphate.

17. A composition according to any one of claims 1-16, for use in increasing the energy capacity within tissue cells.

18. A composition according to any one of claims 1-17, for use in increasing the anaerobic working capacity.

19. A composition according to claim 17 or 18, wherein the subject is human.

20. Use according to claim 21, wherein the subject is male.

21. A method for increasing the anaerobic working capacity wherein a subject is subjected to a building phase and subsequently to a maintenance phase, wherein said building phase comprises intake of a composition comprising creatine, a phosphorus supplement, wherein the phosphorus supplement provides at least 75% of the daily dose value per serving, and a blood buffer, and said maintenance phase comprises intake of said composition, wherein the intake quantity of the composition during maintenance phase is reduced by at least a factor 1.5.